Supplementary Figure S1. New PD-L1 mRNA and protein synthesis are required for IFN-g-induced PD-L1 cell surface expression on melanoma cells.  

A. ActD and CHX completely blocked PD-L1 protein emergence on the surface of melanoma lines after IFN-g exposure. Cultured melanoma cells were preincubated with ActD 10 µg/ml or CHX 2 µg/ml for 1 hr, then incubated with IFN-g 250 U/ml for 6 hr. PD-L1 surface protein was measured by flow cytometry. B. ActD, but not CHX, inhibited new IFN-g-induced PDL1 mRNA transcription. Melanoma lines were treated as described in A. mRNA was measured by qRT-PCR. Ct, cycle threshold. \( \Delta C_t = C_t \text{PDL1} - C_t \text{GAPDH} \) housekeeping gene. The lower the \( \Delta C_t \), the greater PDL1 mRNA expression. As a control, constitutive HLA-DR expression on the same cells was not affected by either ActD or CHX. Representative data from 1 of 2 melanoma cell lines are shown.